

WHAT IS CLAIMED IS:

- 5 1. A non-human transgenic animal whose germ cells and somatic cells contain a knockout mutation in the endogenous $ERR\alpha$ orphan nuclear receptor gene, and wherein said transgenic animal shows a phenotype of an altered fat and/or glucose metabolism as compared to a control animal.
- 10 2. The transgenic animal of claim 1, wherein said germ cells and somatic cells contain a homozygous disruption of said $ERR\alpha$ gene, and wherein said disruption comprises the insertion of a selectable marker sequence.
- 15 3. The non-human transgenic animal of claim 1 or 2, wherein said animal is a mammal.
4. The non-human transgenic animal of claim 3, wherein said animal is a mouse.
- 20 5. The non-human transgenic animal of claims 1 to 4, displaying a lean phenotype.
- 25 6. The non-human transgenic animal of one of claims 1 to 5, whose germ cells and somatic cells additionally comprise a transgene encoding a non endogenous $ERR\alpha$ orphan nuclear receptor gene, wherein said transgene is expressed at levels sufficient to complement the disrupted endogenous $ERR\alpha$ orphan nuclear receptor activity.

7. The non-human transgenic animal of claim 6, wherein said non endogenous $ERR\alpha$ orphan nuclear receptor gene is a human $ERR\alpha$ orphan nuclear receptor gene.

8. The non-human transgenic animal of claim 7, wherein said animal is a mouse and said non-endogenous $ERR\alpha$ is a human $ERR\alpha$ gene.

9. A cell line derived from the non-human transgenic animal of one of claims 1 to 8.

10. A method of producing a non-human transgenic animal, in which at least some cells thereof contain an altered gene encoding an altered $ERR\alpha$, said altered gene having been targeted to disrupt the endogenous $ERR\alpha$ gene in said transgenic animal, said method comprising:

a) providing an altered gene encoding the altered form of $ERR\alpha$ and designed to target and disrupt said endogenous $ERR\alpha$ gene of an embryonic stem cells (ES) of said animal;

b) introducing said altered gene in said ES cells;

c) selecting ES cells in which said altered $ERR\alpha$ gene has disrupted said endogenous $ERR\alpha$ gene;

d) injecting said selected ES cells of c) into blastocysts;

e) implanting said blastocysts of d) in a pseudopregnant animal; and

f) producing a non-human transgenic animal having at least some cells having said altered $ERR\alpha$ gene encoding said altered $ERR\alpha$.

11. The method of claim 10, wherein said non-human transgenic animal is a mouse.

12. A method of producing the non-human transgenic animal of claim 5, said method comprising:

(a) providing a non-human transgenic animal lacking detectable levels of $ERR\alpha$ orphan nuclear receptor gene and exhibiting a lean phenotype;

5 (b) introducing a non endogenous $ERR\alpha$ orphan nuclear receptor transgene encoding a functional $ERR\alpha$ orphan nuclear receptor gene into the pronucleus of a zygote derived from said animal of a), said zygote containing a homozygous disruption of the endogenous $ERR\alpha$ orphan nuclear receptor gene;

10 (c) transplanting said animal zygote into a pseudopregnant compatible animal;

(d) allowing said zygote to develop to term;

(e) obtaining a founder animal carrying said transgene; and

15 (f) breeding said founder animal with a wild-type animal to obtain progeny that express said non endogenous $ERR\alpha$ orphan nuclear receptor transgene at levels sufficient to functionally complement the disrupted $ERR\alpha$ receptor activity.

13. The method of claim 12, wherein said non-human transgenic animal is a mammal.

20 14. The method of claim 12, wherein said mammal is a mouse, and wherein said non-endogenous $ERR\alpha$ transgene is a human $ERR\alpha$ gene.

15. A method for screening and identifying a compound which modulates $ERR\alpha$ orphan nuclear receptor activity, the method including:

25 a) exposing the non-human transgenic animal of one of claims 5 to 7 to a candidate compound, and;

b) determining the activity of said $ERR\alpha$ orphan nuclear receptor in said animal, wherein an increase in the receptor activity as compared to an unexposed

non-human animal is indicative of a compound being capable of increasing $ERR\alpha$ orphan nuclear receptor activity, while a decrease in said receptor activity as compared to an unexposed non-human animal, is indicative of a compound being capable of decreasing $ERR\alpha$ orphan nuclear receptor activity.

- 5 16. The method of claim 15, further comprising a determination of at least one parameter selected from the group consisting of: mass, body temperature, body fat content, fat to lean mass ratio, white adipose tissue deposits, basal metabolic rate, food intake, hepatic synthetic functions, fasting serum triglyceride, serum glucose levels, level of expression of uncoupling protein mRNA in brown
10 adipose tissue (BAT) and skeletal muscle, adipocyte volume in fat pads, lipogenesis, fatty acid esterification and fatty acid oxydation.

17. A method of identifying an agent which modulates fat and/or glucose metabolism *in vivo* comprising:

- 15 a) administering an agent suspected of being a modulator of $ERR\alpha$ activity and/or level in an animal;
 b) measuring lipid and/or glucose levels in the animal of step a) and comparing same with that of a control animal not having been administered said agent, wherein a difference in lipid and/or glucose levels of the animal of step a) as
20 compared to that of the control animal, identifies said agent as a modulator of fat and/or glucose metabolism *in vivo*.

18. Method of identifying an agent which modulates fat and/or glucose metabolism *in vivo* comprising:

- 25 a) providing a promoter operably linked to a selectable or assayable marker, said promoter being modulated by $ERR\alpha$;
 b) measuring or selecting for said marker in a presence and in an absence of an agent suspected of modulating the promoter modulating activity

of $ERR\alpha$, thereby identifying an agent which modulates $ERR\alpha$ activity wherein a difference in the transcriptional activity in the presence of said agent, as compared to that in the absence thereof, identifies said agent as a modulator of $ERR\alpha$ activity;

5 c) administering said agent identified in b) to a non-human transgenic animal according to one of claims 1 to 7; and

 d) measuring lipid and/or glucose levels in said animal of step c) and comparing same with that of a control animal, not having been administered said agent, wherein a difference in lipid and/or glucose levels of the animal of step c) as
10 compared to that of said control animal identifies said agent as a modulator of fat and/or glucose metabolism *in vivo*.

19. The method of claim 18, where the agent is obtained from a library of compounds.

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20. The method of claim 19, wherein the animal is a mammal.

21. The method of claim 20, wherein said mammal is a mouse or human.

20 22. A modulator of fat and/or glucose metabolism *in vivo* identified by any one of the methods of claims 18, 19, 20 or 21.

23. A method of modulating fat tissue growth and/or weight gain, comprising:

25 a) administering to an animal an agent which modulates the promoter activity of a gene, wherein said promoter comprises cis-acting elements selected from the group consisting of:

 i) an estrogen response element;

- ii) TGA AGG TCA;
- iii) AGG TCA NNN TGA CCT; and
- iv) functional variants of i-iii)

5 such as to modulate the level of said gene, thereby modulating fat tissue growth and/or weight gain in said animal.

24. The method of claim 23, wherein said agent modulates said promoter activity of said gene, by decreasing a level and/or activity of $ERR\alpha$.

10 25. The method of claim 24, wherein said agent is an antibody specific to $ERR\alpha$, or an epitope-bearing portion thereof.

26. The method of claim 23, wherein said modulation of said promoter activity is effected by inhibition of $ERR\alpha$ synthesis.

15 27. The method of claim 26, wherein said agent comprises an antisense RNA, complementary to a nucleotide sequence encoding $ERR\alpha$.

20 28. A method of determining whether an agent modulates fat tissue growth and/or weight gain in an animal comprising:

- a) providing a transcriptionally active preparation of $ERR\alpha$ or related factors and a DNA sequence comprising a promoter having a cis-acting sequence which modulates activity thereof by an interaction thereto of said $ERR\alpha$ and related factors;
- 25 b) measuring said transcriptional activity of said promoter or of a binding of at least $ERR\alpha$ or related factors to said cis-acting sequence in a presence and in an absence of an agent suspected of modulating the transcriptional activity of said promoter or the binding of said factors to said cis-

acting sequence, thereby identifying an agent which modulates transcription of said promoter and wherein a difference in the transcriptional activity and/or binding in the presence of said agent, as compared to that in the absence thereof identifies said agent as a modulator of transcription;

- 5 c) administering said agent identified in b) to a non-human transgenic animal according to one of claims 1 to 7; and
- d) measuring fat tissue growth and/or weight gain in the animal of step c) and comparing same with that of a control animal, not having been administered said agent, wherein a difference in fat tissue growth and/or weight
- 10 gain of the animal of step c) as compared to that of the control animal identifies said agent as a modulator of fat tissue growth and/or weight gain *in vivo*.

29. The method of claim 28, where the agent is obtained from a library of compounds.

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30. The method of claim 29, wherein the animal is a mammal.

31. The method of claim 30, wherein said mammal is a mouse or human.

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32. A modulator of fat and/or glucose metabolism *in vivo* identified by any one of the methods of claims 28, 29, 30 or 31.

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33. A method of treating and/or preventing obesity, comprising administering to an obese animal, or an animal susceptible of becoming obese, an agent which modulates the promoter activity of a promoter comprising a cis-acting element selected from the group consisting of:

- i) an estrogen response element;
- ii) TGA AGG TCA;

- iii) AGG TCA NNN TGA CCT; and
- iv) functional variants of I-iii)

wherein cis-acting element is capable of binding to $ERR\alpha$.

5 34. The method of claim 33, wherein said agent reduces the level and/or activity of $ERR\alpha$.

35. A method of determining whether an agent modulates obesity in an animal comprising:

- 10 a) providing a transcriptionally active preparation of $ERR\alpha$ or related factors and a DNA sequence comprising a promoter having a cis-acting sequence which modulates activity thereof by an interaction thereto of said $ERR\alpha$ and related factors;
- 15 b) measuring said transcriptional activity of said promoter or of a binding of at least $ERR\alpha$ or related factors to said cis-acting sequence in a presence and in an absence of an agent suspected of modulating the transcriptional activity of said promoter or the binding of said factors to said cis-acting sequence, thereby identifying an agent which modulates transcription of said promoter and wherein a difference in the transcriptional activity and/or binding
- 20 in the presence of said agent, as compared to that in the absence thereof identifies said agent as a modulator of transcription;
- c) administering said agent identified in b) to a non-human transgenic animal according to one of claims 1 to 7; and
- 25 d) assessing obesity in the animal of step c) and comparing same with that of a control animal, not having been administered said agent, wherein a difference in obesity of the animal of step c) as compared to that of the control animal identifies said agent as a modulator of obesity *in vivo*.

